

# Reproductive Effects of the Pharmaceutical Fluoxetine on Native Freshwater Mussels

Robert B. Bringolf, W. Gregory Cope, Chris Eads, Damian Shea  
North Carolina State University, Raleigh, NC

Rebecca M. Heltsley  
National Institute of Standards and Technology, Charleston, SC

Steve Fraley  
North Carolina Wildlife Resources Commission

Teresa J. Newton  
U.S. Geological Survey, La Crosse, WI



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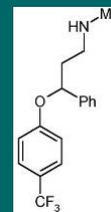
Erin Tracy

## Outline

- Introduction to fluoxetine
- Objectives
- Glochidia release
- Display behavior
- Conclusions
- Implications for future research

## Fluoxetine

- Active ingredient of Prozac™
  - Anti-depressant prescription drug
- Selective Serotonin Reuptake Inhibitor
  - ↑ serotonin in brain
- Pharmaceuticals & personal care products (PPCPs)
- PPCPs increasingly being measured in surface waters
- Benefits are undisputable, but effects on aquatic organisms largely unknown



# Fluoxetine in the environment

- Recently, fluoxetine detected in wastewater effluent
  - up to 0.5 µg/L (Johnson et al. 2005)
- Effects on aquatic organisms?
  - Fluoxetine delays amphibian development (Rogers & Black 2005)
- Serotonin has role in glochidia release
- Serotonin & fluoxetine investigated as control measures for exotic zebra mussels (Fong et al. 1994, 1996)



## Question:

Could fluoxetine disrupt native mussel reproduction?



[dnr.mt.gov/wlr/waterres/mussels/mussel2.htm](http://dnr.mt.gov/wlr/waterres/mussels/mussel2.htm)

## Objectives

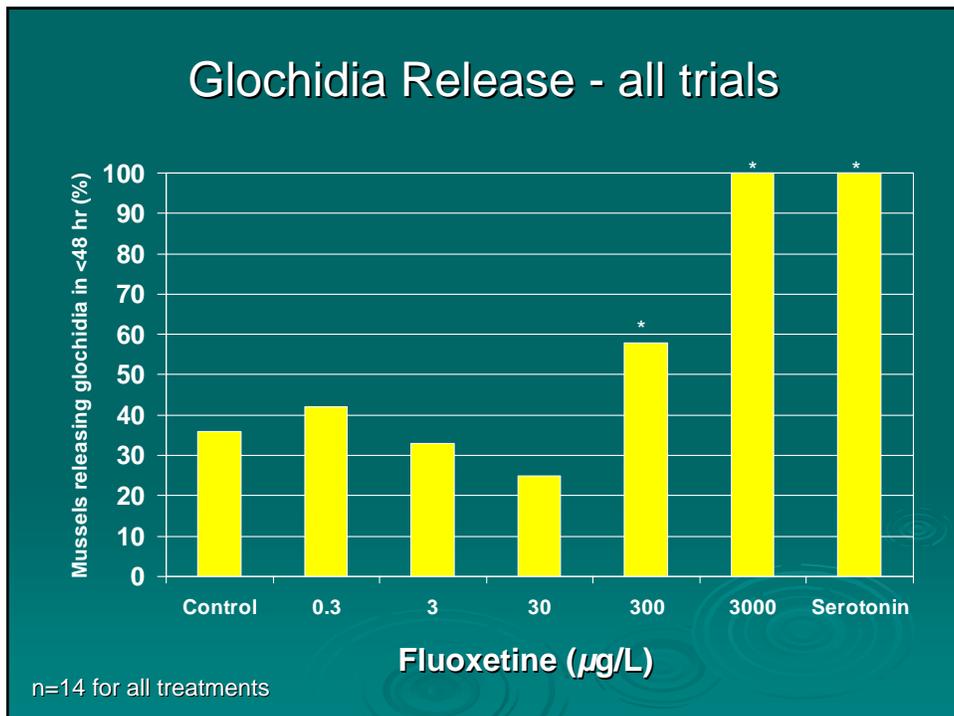
- Evaluate the effects of fluoxetine on glochidia release
- Evaluate effects of fluoxetine on mantle lure display behavior



## Experimental Design – glochidia release

- Adult *Elliptio complanata*
  - gravid females
  - Eno R., Hillsborough, NC
  - Little Cr., Wilson, NC
- 3 trials
  - July '04, June '05, July '06
- Fluoxetine: 0.3 – 3000  $\mu\text{g/L}$ 
  - Static-renewal (24 h)
- Positive control – serotonin
- Endpoints
  - Time to release of glochidia
  - Viability of glochidia released





## Mantle lure display behavior



## Experimental Design – mantle display behavior

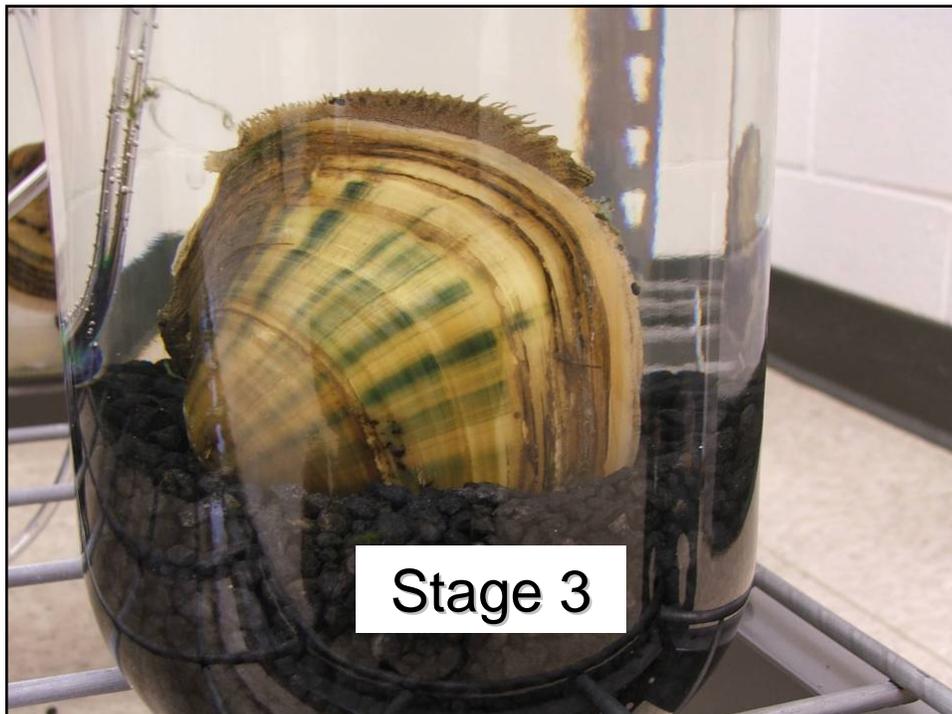
- *Lampsilis cardium*
  - gravid females
  - Upper Mississippi R., Pool 8
- 5 mussels/treatment
- Fluoxetine: 0.3 – 3000  $\mu\text{g/L}$ 
  - Static-renewal (24 h)
- Positive control – serotonin
- Observed daily every 2 hr (10 hr blocks) for 7 d
- Endpoints
  - Release of glochidia
  - Stage of display



## Display stages



Stage	Description
1	Shell closed, no siphoning
2	Siphoning, no mantle visible
3	Partial lure display
4	Full display, no marsupium
5	Full display, marsupium extended
6	Full display, marsupium, lure beating

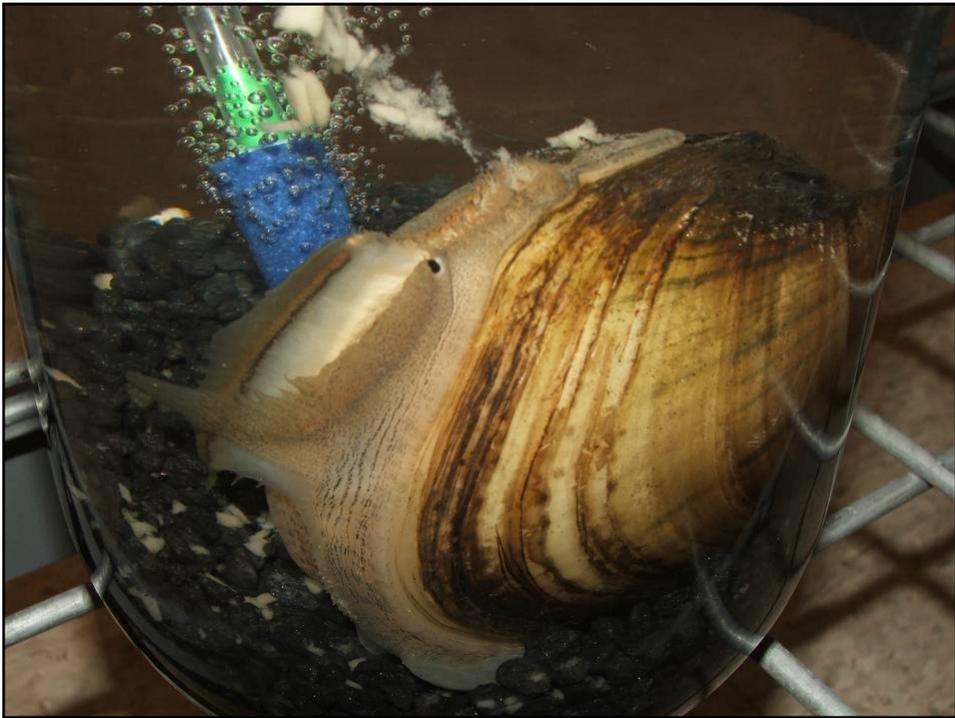


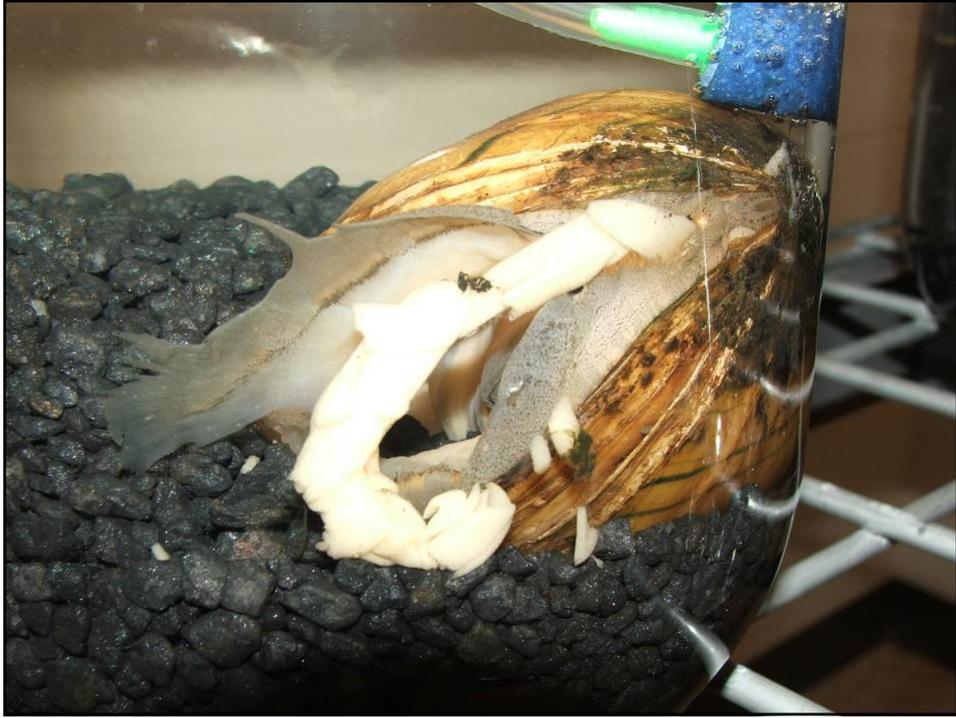
Stage 4



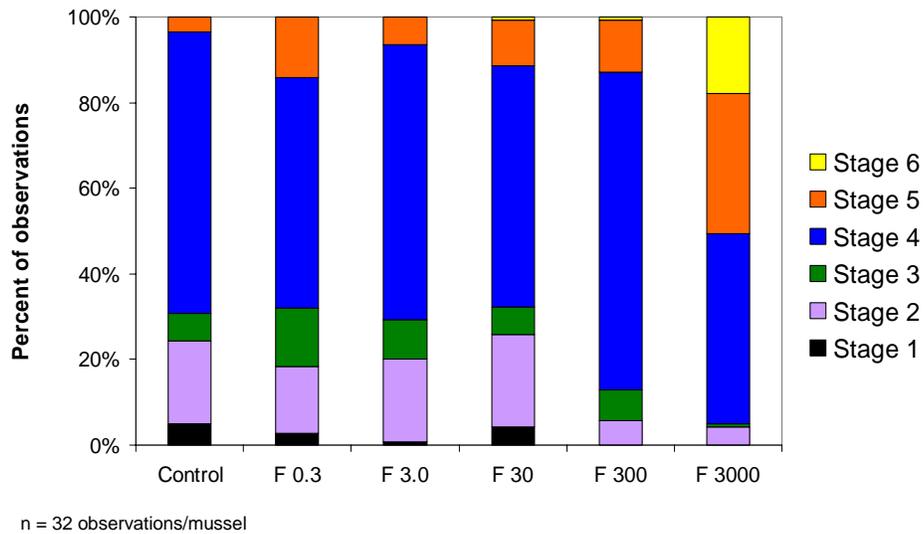
Stage 5







## *L. cardium* display results



## Experimental Design – mantle display behavior

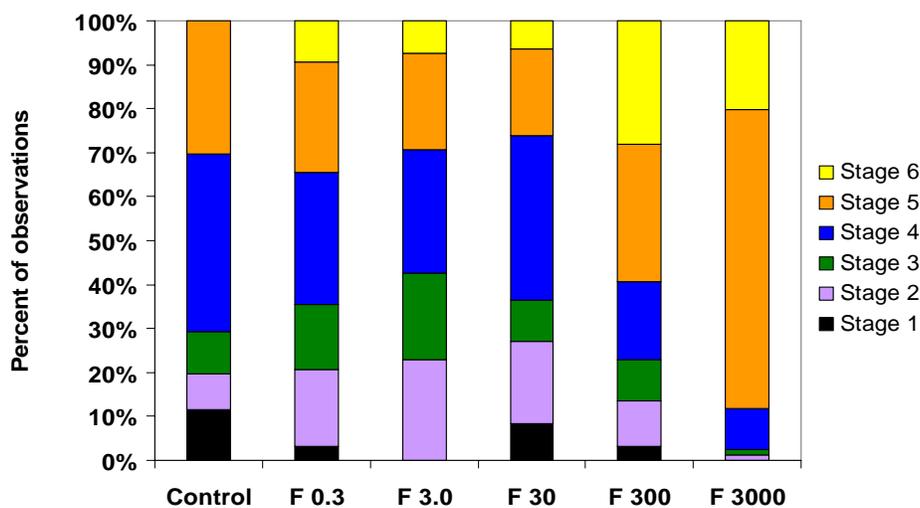
- *Lampsilis fasciola*
  - gravid females
  - Little Tennessee River, Franklin, NC
- 6 mussels/treatment
- Fluoxetine: 0.3 – 3000  $\mu\text{g/L}$
- Positive control – serotonin
- Observed daily every 2 hr (10 hr blocks) for 4 d
- Endpoints
  - Release of glochidia
  - Stage of display
    - Same criteria as *L. cardium*



## *L. fasciola* lure morphology

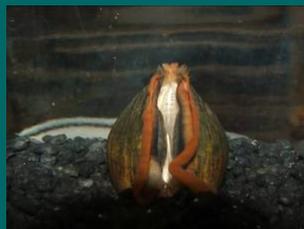


## *L. fasciola* display results



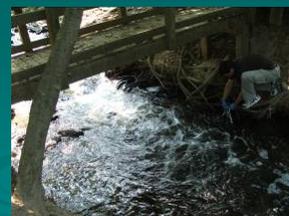
## Behavior test results

- Similar behavioral response for *L. cardium* and *L. fasciola*
  - Significantly greater number in stages 5 & 6 in two highest fluoxetine concentrations
  - No difference in behavior among lure types in *L. fasciola*
- Glochidia release
  - similar to *E. complanata* tests – release at highest fluoxetine concentrations

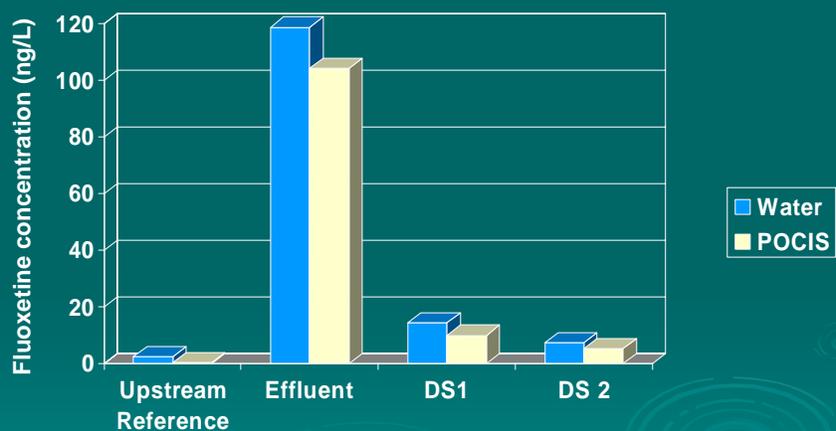


## Environmental samples

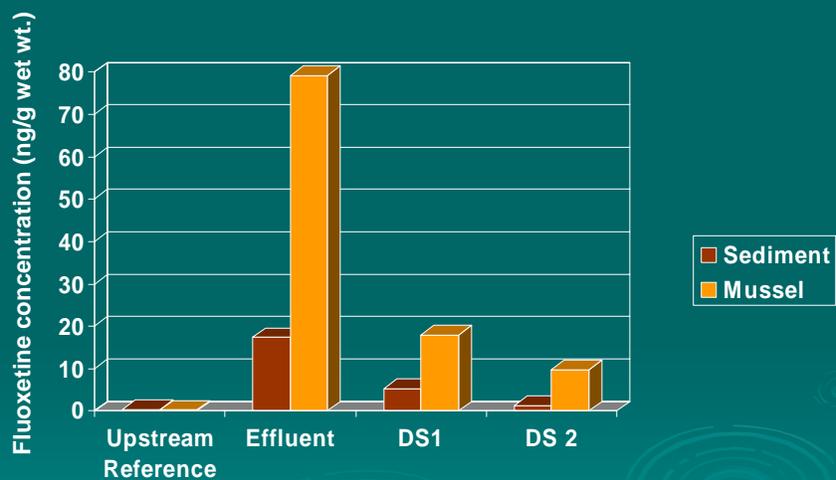
- Fluoxetine (& other SSRIs) in surface water samples
  - Cary, NC 4 sites; near WWTP effluent
  - Grab water samples
  - Passive sampling devices (POCIS)
- Fluoxetine in sediment & mussel tissue
  - Mussels transplanted from clean site to wastewater effluent for 14 d



## Environmental Measurements



## Environmental Measurements



## Summary & Conclusions

- Acute exposure to high concentrations of fluoxetine resulted in premature release of glochidia
  - Mechanistic plausibility for reproductive disruption
- Risk of adverse effects from acute fluoxetine exposure likely low, but effects of chronic exposure to low-levels are not known
- Lure display affected
  - Biological implications?
  - Effects on other behaviors?
- Fluoxetine in water, sediment & mussel tissue

## Things to ponder...

- Freshwater mussels are sentinels of water quality problems
- Fluoxetine is one of hundreds of PPCPs measured in wastewater effluents—chronic, sediment exposures
- Regulatory and management implications?





## Implications



- Population-level consequences of mussel exposure to fluoxetine and other SSRIs may be substantial—requires additional research
- WWTP effluent—downstream concentrations and effects magnified by water reuse
- Ecological Effects – potential for declines and lack of reproduction/recruitment of native fishes, mussels
- Pharmaceutical effects undermining conservation and management efforts???

Mussels on Prozac...